IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	Bey-Dih Chang et al.		
Application No.:	10/801,207	Group:	1633
Filed:	March 16, 2004	Examiner:	Marvich, Maria
Confirmation No.:	3124		
For:	REAGENTS AND METHODS FOR IDENTIFYING AND MODULATING EXPRESSION OF GENES REGULATED BY P21		

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CERTIFICATE OF MAILING

I hereby certify that this correspondence is being deposited with the United States Postal Service with sufficient postage as First Class Mail in an envelope addressed to: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450 on the date indicated below.

Date: _____ Signature_____

Printed Name: _____

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DECLARATION OF IGOR B. RONINSON UNDER 37 C.F.R. §1.132

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

I, Igor B. Roninson, hereby declare as follows.

1. I am a named co-inventor on the above-identified patent application. A copy of my *Curriculum vitae* is attached as Exhibit 1.
2. I have reviewed the Office Action dated January 7, 2009. I understand that claims 1 and 26 of our application are rejected as being anticipated by Fisher and Jiang. In particular, I note that the rejection states that "MDA7 is taught is induced by induction of senescence …, which is also associated with induction of p21 or mda6 … . Identification of an inhibitor of MDA7, through identification of muted MDA7 expression, results in identification of inhibitors of p21 and senescence inherently." For the reasons set forth

below, I do not agree that identification of an inhibitor of MDA7, through identification of muted MDA7 expression, results in identification of inhibitors of p21 and senescence inherently.

3. Our 2002 article (Chang,B.D., Swift,M.E., Shen,M., Fang,J., Broude,E.V., and Roninson,I.B. (2002). Molecular determinants of terminal growth arrest induced in tumor cells by a chemotherapeutic drug. Proc. Natl. Acad. Sci. U. S. A. *99*, 389-394.) addresses the question of which of the genes that are induced or repressed in senescent cells, along with p21 induction, change their expression because of p21 induction. Our study demonstrates that many of the genes that are induced in senescent HCT116 cells are induced independently of p21, although p21 is also induced in these cells. Such p21-independent senescence-associated genes would not constitute reporters claimed under our invention.
4. Fischer and Jiang demonstrate that the gene that they termed mda-7, and which is now known as IL24, is induced in melanoma cells undergoing terminal differentiation and growth arrest (they never show or suggest senescence), along with p21 (mda-6). Fischer and Jiang provide no evidence and do not even suggest that mda-7/IL24 is induced by p21. Hence, there is no reason to claim that mda-7/IL24 anticipates our reporter genes.
5. I can also add that I checked the results of our unpublished microarray analysis of the effect of IPTG-inducible p21 on the expression of all the known genes in HT1080 cells, with regard to the effects of p21 on IL24. p21 did not induce IL24 in HT1080 cells.
6. I hereby further declare that all statements made herein of my own knowledge are true and all statements made on information and belief are believed to be true and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

So made by me this 4th date of May 2009.

/Igor B. Roninson/

Igor B. Roninson

BIOGRAPHICAL SKETCH

Provide the following information for the key personnel and other significant contributors.
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NAME	POSITION TITLE
Igor B. Roninson	Director of Cancer Center, Ordway Research Institute
eRA COMMONS USER NAME RONINSON	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, and include postdoctoral training.)*

INSTITUTION and LOCATION	DEGREE *(if applicable)*	YEAR(s)	FIELD OF STUDY
Moscow University, Moscow, Russia	M.S.	1977	Virology
Massachusetts Institute of Technology	Ph.D.	1982	Biochemistry
Massachusetts Institute of Technology	Postdoc.	1982-1984	Molecular Biology

A. Positions and Honors

Positions and employment

1979 Laboratory Scientist, Department of Biological Chem., Univ. of Maryland School of Medicine at Baltimore.
1979-82 Predoctoral Trainee, Department of Biology, Massachusetts Institute of Technology.
1982-84 Postdoctoral Fellow, Department of Biology, Massachusetts Institute of Technology.
1984-89 Assistant Professor, Department of Genetics, University of Illinois at Chicago.
1989-93 Associate Professor, Department of Genetics, University of Illinois at Chicago.
1993-1995 Professor, Department of Genetics, University of Illinois at Chicago.
1994-2003 Head, Division of Molecular Oncology, Dept. of Molecular Genetics, University of Illinois at Chicago.
1995-2003 Distinguished University Professor, Department of Molecular Genetics, University of Illinois at Chicago.
From 2003 Director, Cancer Center, Ordway Research Institute, Albany, NY
From 2004 Research Professor of Biology, Rensselaer Polytechnic Institute, Troy, NY
From 2004 Adjunct Professor of Biology, State University of New York at Albany, Albany, NY
From 2005 Adjunct Professor of Medicine, Albany Medical College, Albany, NY

Other Experience and Honors:

Editorial Boards: (present) Cancer Res., Cell Cycle, Aging, Drug Resistance Updates, Cancer Letters; (past) J. Natl. Cancer Inst., Anticancer Drugs, Cancer and Metastasis Rev.
Invited speaker at 83 meetings in 20 countries - 1985-2008.
Inventor of 38 issued US patents.
Editor, *"Molecular and Cellular Biology of Multidrug Resistance in Tumor Cells"*, Plenum Press, NY, 1991.
Editor, *"Beyond Apoptosis: Cellular Outcomes of Cancer Therapy"*, Informa Healthcare, NY, 2008.
Awards: Honors Diploma, Moscow State University (1977); Postdoctoral Fellowship, American Cancer Society (1982); University Scholar Award, University of Illinois (1987); Faculty Research Award, American Cancer Society (1989); MERIT Award, National Cancer Institute (1993); Rhoads Award for Meritorious Achievement in Cancer Research, AACR (1994); Distinguished Lecturer in Pharmacology, St. Jude's Children's Research Hospital (1995); UICC Roll of Honor (1999); Life Extension Prize, Regenerative Medicine Secretariat (2000); Inventor of the Year Award, University of Illinois (2000); Sagov-Pomereniec Lectureship, Hebrew University, Israel (2001).

B. Selected Publications (in chronological order from a total of 151):

Roninson, I.B., Abelson, H., Housman, D.E., Howell, N., and Varshavsky, A. (1984). Amplification of specific DNA sequences correlates with multidrug resistance in Chinese hamster cells. *Nature* 309, 626-628.

Roninson, I.B., Chin, J.E., Choi, K., Gros, P., Housman, D.E., Fojo, A., Shen, D., Gottesman, M.M., and Pastan, I. (1986). Isolation of human *mdr* DNA sequences amplified in multidrug-resistant KB carcinoma cells. *Proc. Natl. Acad. Sci. USA* 83, 4538-4542.

Chen, C.-j., Chin, J.E., Ueda, K., Clark, D.P., Pastan, I., Gottesman, M.M., and Roninson, I.B. (1986). Internal duplication and homology with bacterial transport proteins in the *mdr*1 (P-glycoprotein) gene from multidrug-resistant human cells. *Cell* 47, 381-389.

Choi, K., Chen, C.-j., Kriegler, M., and Roninson, I.B. (1988). An altered pattern of cross-resistance in multidrug-resistant human cells results from spontaneous mutations in the *mdr*1 (P-glycoprotein) gene. *Cell* 53, 519-529.

Fukumoto, M., Shevrin, D.H., and Roninson, I.B. (1988). Analysis of gene amplification in human tumor cell lines. *Proc. Natl. Acad. Sci. USA* 85, 6846-6850.

Noonan, K.E., Beck, C., Holzmayer, T. A., Chin, J.E., Wunder, J.S., Andrulis, I.L., Gazdar, A.F., Willman, C.L., Griffith, B., Von Hoff, D.D., and Roninson, I.B. (1990). Quantitative analysis of *MDR*1 (multidrug resistance) gene expression in human tumors by polymerase chain reaction. *Proc. Natl. Acad. Sci. USA* 87, 7160-7164.

Chaudhary, P.M. and Roninson, I.B. (1991). Expression and activity of P-glycoprotein, a multidrug efflux pump, in human hematopoietic stem cells. *Cell* 66, 85-94.

Choi, K., Frommel, T.O., Kaplan Stern, R., Perez, C.F., Kriegler, M., Tsuruo, T., and Roninson, I.B. (1991). Multidrug resistance after retroviral transfer of the human *MDR*1 gene correlates with P-glycoprotein density in the plasma membrane and is not affected by cytotoxic selection. *Proc. Natl. Acad. Sci. USA*, 88, 7386-7390.

Holzmayer, T.A., Pestov, D.G., and Roninson, I.B. (1992). Isolation of dominant negative mutants and inhibitory antisense RNA sequences by expression selection of random DNA fragments. *Nucleic Acids Res.* 20, 711-717.

Mechetner, E.B. and Roninson, I.B. (1992). Efficient inhibition of P-glycoprotein-mediated multidrug resistance with a monoclonal antibody. *Proc. Natl. Acad. Sci. USA* 89, 5824-5828.

Gudkov, A.V., Zelnick, C., Kazarov, A.R., Thimmapaya, R., Suttle, D.P., Beck, W.T., and Roninson, I.B. (1993). Isolation of genetic suppressor elements, inducing resistance to topoisomerase II-interactive cytotoxic drugs, from human topoisomerase II cDNA. *Proc. Natl. Acad. Sci. USA* 90, 3231-3235.

Gudkov, A.V., Kazarov, A.R., Thimmapaya, R., Axenovich, S., Mazo, I., and Roninson, I.B. (1994). Cloning mammalian genes by expression selection of genetic suppressor elements: association of kinesin with drug resistance and cell immortalization. *Proc. Natl. Acad. Sci. USA*, 91, 3744-3748.

Roninson, I.B., Gudkov, A.V., Holzmayer, T.A., Kirschling, D.J., Kazarov, Kazarov, A.R., Zelnick, C.R., Mazo, I.A., Axenovich, S., and Thimmapaya, R. (1995). Genetic suppressor elements: new tools for molecular oncology: 13th Cornelius P. Rhoads Memorial Award lecture. *Cancer Res.* 55, 4023-4028.

De Graaf, D., Sharma, R.C., Mechetner, E.B., Schimke, R.T., and Roninson, I.B. (1996). P-glycoprotein confers methotrexate resistance in 3T6 cells with deficient carrier-mediated methotrexate uptake. *Proc. Natl. Acad. Sci. USA*, 93, 1238-1242.

Mechetner, E.B., Schott, B., Morse, B.S., Stein, W.D., Druley, T., Davis, K., Tsuruo, T. and Roninson, I.B. (1997). P-glycoprotein function involves conformational transitions detectable by differential immunoreactivity. *Proc. Natl. Acad. Sci.* 94, 12908-12913.

Gudkov, A.V., Roninson, I.B. and Brown, R. (1999). Functional approaches to gene isolation in mammalian cells. *Science* 285, 299a.

Chang, B.D., Broude, E.V., Dokmanovic, M., Zhu, H., Ruth, A., Xuan, Y., Kandel, E.S., Lausch, E., Christov, K. and Roninson, I.B. (1999). A senescence-like phenotype distinguishes tumor cells that undergo terminal proliferation arrest after exposure to anticancer agents. *Cancer Res.* 59, 3761-3767.

Chang, B.D., Watanabe, K., Broude, E.V., Fang, J., Poole, J.C., Kalinichenko, T.V., and Roninson, I.B. (2000). Effects of $p21^{Waf1/Cip1/Sdi1}$ on Cellular Gene Expression: Implications for Carcinogenesis, Senescence and Age-Related Diseases. *Proc. Natl. Acad. Sci. USA* 97, 4291-4296.

Chang, B.D., Broude, E.V., Fang, J., Kalinichenko, T.V., Abdryashitov, R., Poole, J.C., and Roninson, I.B. (2000). $p21^{Waf1/Cip1/Sdi1}$-induced growth arrest is associated with depletion of mitosis-control proteins and leads to abnormal mitosis and endoreduplication in recovering cells. *Oncogene* 19, 2165-2170.

Ruth, A. and Roninson, I.B. (2000). Effects of the multidrug transporter P-glycoprotein on cellular responses to ionizing radiation. *Cancer Res.*, 60, 2576-2579.

Levenson, V.V., Davidovich, I.A. and Roninson, I.B. (2000). Pleiotropic resistance to DNA-interactive drugs is associated with increased expression of genes involved in DNA replication, repair and stress response. *Cancer Res.* 60, 5027-5030.

Druley, T.E., Stein, W.D., and Roninson, I.B. (2001). Analysis of MDR1 P-glycoprotein conformational changes in permeabilized cells using differential immunoreactivity. *Biochemistry* 40, 4312-4322.

Druley, T.E., Stein, W.D., Ruth, A., and Roninson, I.B. (2001). P-glycoprotein-mediated colchicine resistance in different cell lines correlates with the effects of colchicine on P-glycoprotein conformation. *Biochemistry* 40, 4323-4331.

Ruth, A., Stein, W.D., Rose, E., and Roninson, I.B. (2001). Coordinate changes in drug resistance and drug-induced conformational transitions in altered-function mutants of the multidrug transporter P-glycoprotein. *Biochemistry* 40, 4332-4339.

Kramer, D.L., Chang, B.D., Chen, Y., Diegelman, P., Alm, K., Roninson, I.B., and Porter, C.W. (2001).

Polyamine depletion in human melanoma cells leads to G_1 arrest associated with induction of $p21^{WAF1/CIP1/SDI1}$, changes in the expression of p21-regulated genes, and a senescence-like phenotype. *Cancer Res.* 61, 7754-7762.

Chang, B.D., Swift, M.E., Shen, M., Fang, J., Broude, E.V., and Roninson, I.B. (2002). Molecular determinants of terminal growth arrest induced in tumor cells by a chemotherapeutic drug. *Proc. Natl. Acad. Sci. USA* 99, 389-394.

Dokmanovic, M., Chang, B.D., Fang, J., and Roninson, I.B. (2002). Retinoid-induced growth arrest of breast carcinoma cells involves co-induction of multiple growth-inhibitory genes. *Cancer Biol. Ther.* 1, 16-19.

Zhu, H., Chang, B.D., Uchiumi, T., and Roninson, I.B. (2002). Identification of promoter elements responsible for transcriptional inhibition of Polo-like kinase 1 and Topoisomerase IIα genes by $p21^{WAF1/CIP1/SDI1}$. *Cell Cycle* 1, 59-66.

Roninson, I.B. (2002). Oncogenic functions of tumor suppressor $p21^{Waf1/Cip1/Sdi1}$: association with cell senescence and tumor-promoting activities of stromal fibroblasts. *Cancer Lett.*, 179, 1-14.

Roninson, I.B. and Gudkov, A.V. (2002). Genetic suppressor element (GSE) methodology and its applications to characterization and identification of tumor suppressor genes. In: *Methods in Molecular Medicine: Analysis of Tumor Suppressor Genes, volume I* (W.S. El-Deiry, Ed.), Humana Press, pp. 411-434.

Roninson, I.B. (2002). Tumor senescence as a determinant of drug response *in vivo*. *Drug Resistance Updates* 5, 204-208.

Gregory, D.J., Garcia-Wilson, E., Poole, J.C., Snowden, A.W., Roninson, I.B., and Perkins, N.D. (2002). Induction of transcription through the p300 CRD1 motif by $p21^{WAF1/CIP1}$ is core promoter specific and cyclin dependent kinase independent. *Cell Cycle* 1, 343-350.

Roninson, I.B. and Dokmanovic, M. (2003). Induction of senescence-associated growth inhibitors in the tumor-suppressive function of retinoids. *J. Cell. Biochem.* 88, 83-94.

Roninson, I.B. (2003). Tumor cell senescence in cancer treatment. *Cancer Res.* 11, 2705-2715.

Primiano, T., Baig, M., Maliyekkel, A., Chang, B.D., Fellars, S., Sadhu, J., Axenovich, S., Holzmayer, T.A. and Roninson, I.B. (2003). Identification of potential anticancer drug targets through the selection of growth-inhibitory genetic suppressor elements. *Cancer Cell*, 4, 41-53.

Shay, J.W. and Roninson, I.B. (2004). Hallmarks of senescence in carcinogenesis and cancer therapy. *Oncogene*, 23, 2919-2933.

Nickoloff, B.J., Lingen, M.W., Chang, B.D., Shen, M., Swift, M., Curry, J., Bacon, P., Bodner, B., and Roninson, I.B. (2004). Tumor suppressor maspin is upregulated during keratinocyte senescence, exerting a paracrine anti-angiogenic activity. *Cancer Res.* 64, 2956-2961.

Poole, J.C., Thain, A., Perkins, N.D., and Roninson, I.B. (2004). Induction of transcription by $p21^{Waf1/Cip1/Sdi1}$: role of NFκB and effect of non-steroidal anti-inflammatory drugs. *Cell Cycle* 3, 931-940.

Roninson, I.B. (2005). Seeking favors from Nature. *Cancer Biol. Ther.* 4, 794-799.

Vigneron, A., Roninson, I.B., Gamelin, E.,and Coqueret, O. (2005). Src inhibits Adriamycin-induced senescence and G2 checkpoint arrest by blocking the induction of p21waf1. *Cancer Res.* 65, 8927-8935.

Currid, C.A., O'Connor, D.P., Chang, B.D., Gebus, C., Harris, N., Dawson, K.A., Dunn, M.J., Pennington, S.R., Roninson, I.B., Gallagher, W.M. (2006). Proteomic analysis of factors released from p21-overexpressing tumour cells. *Proteomics,* 6, 3739-3753.

Chen, Y., Dokmanovic, M., Stein, W.D., Ardecky, R.J., Roninson, I.B. (2006). Agonist and antagonist of retinoic acid receptors cause similar changes in gene expression and induce senescence-like growth arrest in MCF-7 breast carcinoma cells. *Cancer Research,* 66, 8749-8761.

Maliyekkel, A., Davis, B. M., and Roninson, I.B. (2006). Cell cycle arrest drastically extends the duration of gene silencing after transient expression of short hairpin RNA. *Cell Cycle* 5, 2390-2395.

Shtutman, M., Levina, E., Ohouo, P., Baig, M., and Roninson, I.B. (2006). Cell adhesion molecule L1 disrupts E-cadherin containing adherens junctions and increases scattering and motility of MCF7 breast carcinoma cells. *Cancer Research*, 66, 11370-11380.

Barre, B., Vigneron, A., Perkins, N., Roninson, I.B., Gamelin, E., and Coqueret, O. (2007). The STAT3 oncogene as a predictive marker of drug resistance. *Trends Mol Med.* 13, 4-11.

Broude, E.V., Swift, M.E., Vivo, C., Chang, B.-D., Davis, B.M., Kalurupalle, S., Blagosklonny, M.V., and Roninson, I.B. (2007). $p21^{Waf1/Cip1/Sdi1}$ mediates retinoblastoma protein degradation. *Oncogene* 26, 6954-6958.

Broude, E.V., Demidenko, Z.N., Vivo, C., Swift, M.E., Davis, B.M., Blagosklonny, M.V., and Roninson, I.B. (2007). p21 (CDKN1A) is a negative regulator of p53 stability. *Cell Cycle* 6,1468-1471.

Shao, Y., Chan, C.Y., Maliyekkel, A., Lawrence, C.E., Roninson, I.B., and Ding. Y. (2007). Effect of target

secondary structure on RNAi efficiency. *RNA* 13, 1631-1640.
Broude, E.V., Loncarek, J., Wada, I., Cole, K., Hanko, C., Swift, M. and Roninson, I.B. (2008). Mitotic catastrophe in cancer therapy. In "Beyond Apoptosis: How Anticancer Agents Stop the Growth of Tumor Cells", I.B. Roninson, J.M. Brown and D.E. Bredesen, Editors, Informa Healthcare, pp.307-320.
Roninson, I.B. and Broude, E.V. (2008). Treatment-induced tumor cell senescence and its consequences. In "Beyond Apoptosis: How Anticancer Agents Stop the Growth of Tumor Cells", I.B. Roninson, J.M. Brown and D.E. Bredesen, Editors, Informa Healthcare, pp. 223-249.
Broude, E.V., Loncarek, J., Wada, I., Cole, K., Hanko, C., and Roninson, I.B. (2008). Fluorescent and phase contrast video microscopy of mitotic catastrophe in irradiated tumor cells. DVD supplement to "Beyond Apoptosis: How Anticancer Agents Stop the Growth of Tumor Cells", I.B. Roninson, J.M. Brown and D.E. Bredesen, Editors, Informa Healthcare.
Yates, K.E., Korbel, G.A., Shtutman, M., Roninson, I.B., and DiMaio, D. (2008). Repression of the SUMO-specific protease Senp1 induces p53-dependent premature senescence in normal human fibroblasts. *Aging Cell* 7, 609-621.
Tapias, A., Ciudad, C.J., Roninson, I.B. and Noé, V. (2008). Regulation of SP1 by cell cycle related proteins. *Cell Cycle* 7, 2856-2867.
Chan, C.Y., Carmack, C.S., Long, D., Maliyekkel, A., Shao, Y, Roninson, I.B., and Ding, Y. (2008). A structural interpretation of the effects of GC-content on efficiency of RNA interference. *BMC Bioinformatics* 10, Suppl 1:S33.
Gravina, S., Lescai, F., Hurteau, G., Brock, J.B., Saramaki, A., Salvioli, S., Franceschi, C., and Roninson, I.B. (2009). Identification of single nucleotide polymorphisms in the p21 (CDKN1A) gene and correlations with longevity in the Italian population. *Aging*, in press.

C. Research Support

Ongoing Support

NIH R01AG028687 09/30/2007-09/29/2012 Role: Principal Investigator (PI)
Project Title: Secretory Patterns of Senescent Tumor Cells.
Project Goals: The goals are to characterize the spectrum of paracrine growth-regulatory activities of tumor cells rendered senescent by different types of anticancer agents.

Veterans Affairs Merit Review Entry Program Grant; Mian, B. (P.I.) 04/01/2006-03/31/2009 Role: Mentor
Project Title: Senescence-Associated Growth-Regulatory Proteins in Prostate Cancer.
Project Goals: This medical scientist training grant for a urological oncologist, mentored by the P.I., analyzes the expression of growth-regulatory proteins produced by senescent cells in clinical and experimental prostate cancer.

BiPar Sciences, Inc. contract 09/01/2006-12/31/2008 Role: Principal Investigator (PI)
Project Title: Cell cycle activities of newly-synthesized experimental compounds
Project goals: To characterize the effects of several experimental compounds in human cancer cell lines.

Recently Completed Support

NIH/NCI RO1 CA89636 06/01/2001-05/31/2008 Role: Principal Investigator (PI)
Project Title: Accelerated Senescence in Tumor Cells
Project Goals: The goals are to identify genes that show altered expression in tumor cells undergoing drug-induced accelerated senescence, investigate the role of such genes in senescence-associated growth arrest, and identify promoter sites responsible for transcriptional changes of these genes.

NIH RO1 AG17921 04/01/2002-03/31/2008 Role: Principal Investigator (PI)
Project Title: p21 as an Inducer of Disease-Associated Gene Expression Role: Principal Investigator (PI)
Project Goals: The goals are to characterize the mechanisms of the induction of different disease-associated genes by p21, in normal and transformed human fibroblasts.

NIH RO1 CA95727 04/01/2002-03/31/2008 Role: Principal Investigator (PI)
Project Title: Mitotic Catastrophe in Tumor Cells
Project Goals: The goals are to characterize the mechanisms of mitotic catastrophe induced in tumor cells by ionizing radiation or by CDK inhibitors p21 or p16, and to investigate the role of mitotic catastrophe in the antiproliferative effect of anticancer agents.

NIH R33 CA95996 09/30/2003-09/29/2007 Role: Principal Investigator (PI)
Project Title: Function-based selection of target genes in tumor cells.
Project Goals: Goals include identification of genes essential for the growth of different tumor cell types through expression selection of growth-inhibitory Genetic Suppressor Elements (GSEs).